BEET ROOT POMACE - A GOOD SOURCE OF ANTIOXIDANT PHYTOCHEMICALS

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ABSTRACT

The present study describes the *in vitro* antioxidant activity of ethanol extracts of beet root pomace Total content of phenolics, flavonoids and betalains were determinated after solid-phase extraction. Evaluation of antioxidative activity of beet root pomace extract against stable 1,1-diphenyl-2-picrylhydrazyl (DPPH) radicals was determined spectofotometrically and against reactive hydroxil radicals by electron spin resonance (ESR) spectroscopy. Also, antioxidant activity on hydroxyl radicals after *in vitro* incubation in stimulated stomach model system was observed.

1. INTRODUCTION

Processing of fruits and vegetables results in high amounts of waste materials (by-products) which are economical and ecological deficit problem. These products are also promising sources of bioactive antioxidants and color giving compounds, which could be used as additives in food, pharmaceutical and cosmetic industry. In numerous diets, beet root is a very significant part and represent an important source of bioavailable compounds such as polyphenolic compounds, carotenoids, betalains, vitamins and mineralals. The beet root pomace, waste product generated primarily during juice processing, is a potential source of natural antioxidant compounds for use as dietary or food antioxidant.

2. MATERIAL AND METHODS

Beet root was pulped using a fruit mill and beet root pomace was used in this work.
Sample of beet root pomace (100 g) was extracted with 50% ethanol (1000ml) containing 0,5% acetic acid for 30min. The obtained extract was evaporated to dryness under reduced pressure (m = 8,6661g).
Extract was purified using solid-phase extraction (SPE) with CHROMABOND C_{18} columns. The purified beet root extract was evapored to dryness under reduced pressure (m = 0.1109 g).
Total phenolic compounds in extract were determined spectrophotometrically using the Folin-Ciocalteu reagent (expressed as mg chlorogenic acid equivalents per g dry weight of beet root pomace).
Total flavonoids in extract were estimated spectrophotometrically according to Markham (expressed as mg rutin per g dry weight of beet root pomace).
Total betalain contents (betacyanins and betaxanthins) were measured spectrophotometrically using von Elbe method (expressed as mg betacyanins and betaxanthins per g dry weight of beet root pomace).
Content of phenolic acids and flavonoids was quantified by HPLC analysis. Instrumentation and chromatographic conditions HPLC analysis was performed using a liquid chromatograph HPLC Agilent 1200 equipped with a DAD (Diode Array Detector). A Agilent column, Eclipse XDB-C18, 1,8 μm, 4,6 x 50 mm, was used at a flow-rate of 1

ml/min. Solvent gradient was performed by varying the proportion of solvent A (methanol) to solvent B (1% HCOOH in water). The spectra were acquired in the range 190–400 nm.

Antioxidant activity against stable 1,1-diphenyl-2-picrylhydrazyl (DPPH) radicals was determined spectofotometrically. The hydrogen atom or electron donation abilities of the extract was measured from the bleaching of a purple-colored methanol solution of stable DPPH radical. The influence of different ethanol extract on the formation and stabilization of hydroxyl radicals in Fenton reaction system was investigated using ESR spin-trapping method. The Fenton reaction was conducted by mixing 0.2 mL of 0.3 M DMPO, 0.2 mL of 10 mM H_2O_2, and 0.2 mL of 10 mM Fe^{2+} (control). ESR spectra were recorded after 5 minutes.
Also, antioxidant activity of enriched beet root juice on hydroxyl radicals after *in vitro* incubation in stimulated stomach model system was observed.

3. RESULTS AND DISCUSSION

Table 1. Antioxidant compounds in beet root pomace extract

Antioxidant compounds		mg/g
Total phenolic compounds		117,863
Total flavonoids		54,844
Total betalains	Betacyanins	14,129
	Vulgaxanthins	8,324

Table 2. Content of phenolic acids and flavonoids quantified by HPLC analysis

Phenolic acids and flavonoids	mg/g
Catechin	37,497
Vanilic acid	1,205
Protocatechin	0,598
Cimet acid	0,015
Quercetin	0,006

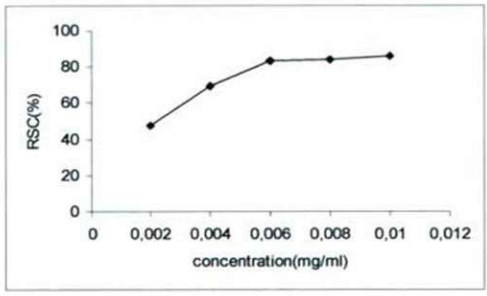

Figure 2. Antioxidant activity of beet root pomace extract against stable DPPH radicals

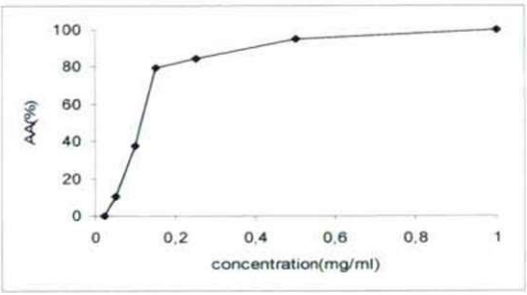

Figure 3. Antioxidant activity of beet root pomace extract against reactive hydroxil radical

Results show that beet root pomace possess considerable amounts of phenolic compounds and betalains. HPLC analysis showed that the most abundant flavonoid was catechin (37,497 mg/g), probably the most responsible component for strong antioxidant activity of extract. The scavenging activity on hydroxyl radicals was increased in the presence of different amounts of beet root extracts.The highest investigated concentration (1 mg/ml) of ethanol extract inhibits completely the formation of hydroxyl radical. The capacity of beet root pomace to inhibit hydroxyl radical generated by the Fenton reaction could be due to direct scavenging effect and/or to inhibit of hydroxyl generation. The second mechanism occurs by ion chelation. Antioxidant activity against hydoxyl radicals of enriched juice was AA_{OH}=69,38%, and after *in vitro* incubation in simulated stomach model system was AA_{OH}=92,34%.

4. CONCLUSION:

Ethanol extract of beet root pomace has strong antioxidant activity and possess considerable amounts of phenolic compounds and betalains and significant radical scavenging activity on stable DPPH (IC_{50}^{DPPH} was 0,002 mg/ml) and highly reactive hydroxyl radicals (IC_{50} was 0,115 mg/ml). Also, the increase of antioxidant activity on hydroxyl radicals after the *in vitro* incubation in simulated stomach model system suggested the increase of bioavailability of natural antioxidants present in extract. These results show that beet root pomace, waste material after juice processing can be used as a good source of phytochemicals and as a natural additive or functional foods.

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